

**TOWNSEND**  
*and*  
**TOWNSEND**  
*and*  
**CREW**

LLP

JC14 Rec'd PCT/PTO  
Denver, CO  
Tel 303 571-4000

10/528747  
22 MAR 2005

San Francisco

Palo Alto, California  
Tel 650 326-2400

Seattle, Washington  
Tel 206 467-9600

Two Embarcadero Center  
Eighth Floor  
San Francisco  
California 94111-3834  
Tel 415 576-0200  
Fax 415 576-0300

May 7, 2004

**VIA EXPRESS MAIL, WITH RETURN POSTCARD ENCLOSED**

PCT International Application Processing Div.  
USPTO International Division  
Assistant Commissioner for Patents  
Mail Stop PCT  
P.O. Box 1450  
Alexandria, VA 22313-1450

Re: International Application No. PCT/US2003/032086  
Applicants: THE GOVERNMENT OF THE USA AS REPRESENTED BY THE  
SECRETARY OF THE DEPT. OF HEALTH AND HUMAN SERVICES et al.  
Inventors: SHOEMAKER et al.  
Filed: 08 October 2003  
Express Mail Label No.: EV 383 385 199 US  
Date of Mailing: 07 May 2004  
Our File No.: 15280-4621PC

Dear Officer:

Enclosed are the Chapter II Demand and substitute page 24 of the specification submitted as an Amendment under Article 34. Enclosed also is a redlined version of the substitute page.

It is our belief that the above change does not include matter which go beyond the disclosure in the international application as filed.

Thank you for your attention to this matter.

Respectfully submitted,

  
Jean M. Lockyer, Ph.D.  
Patent Agent

JML/e1q

Enclosures: Chapter II Demand  
Substitute page 24 of the Specification  
Redlined Version of Substitute Page  
Sequence Listing with Diskette  
Transmittal Letter  
Postcard

The demand must be filed directly with the competent International Preliminary Examining Authority, if two or more Authorities are competent, with the one chosen by the applicant. The full name or two-letter code of that Authority may be indicated by the applicant on the line below:

IPEA/ US

# PCT

## DEMAND

### CHAPTER II

under Article 31 of the Patent Cooperation Treaty:

The undersigned requests that the international application specified below be the subject of international preliminary examination according to the Patent Cooperation Treaty.

For International Preliminary Examining Authority use only

Identification of IPEA		Date of receipt of DEMAND	
<b>Box No. I IDENTIFICATION OF THE INTERNATIONAL APPLICATION</b>		Applicant's or agent's file reference 15280-4621PC	
International application No. PCT/US2003/032086	International filing date (day/month/year) 08 October 2003 (08.10.03)	(Earliest) Priority date (day/month/year) 08 October 2002 (08.10.02)	
Title of invention IDENTIFICATION OF ANTI-HIV COMPOUNDS INHIBITING VIRUS ASSEMBLY AND BINDING OF NUCLEOCAPSID PROTEIN TO NUCLEIC ACID			
<b>Box No. II APPLICANT(S)</b>			
Name and address: (Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)  THE GOVERNMENT OF THE USA AS REPRESENTED BY THE SECRETARY OF THE DEPT. OF HEALTH AND HUMAN SERVICES OTT-NIH 6011 Executive Boulevard, Room 325 Rockville, MD 20852 United States of America		Telephone No.: (301) 496-7056	
		Facsimile No.: (301) 402-0220	
		Teleprinter No.:	
		Applicant's registration No. with the Office 44,879	
State (that is, country) of nationality: US		State (that is, country) of residence: US	
Name and address: (Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)  SHOEMAKER, Robert H. 22606 Peach Tree Road Boysds, MD 20841 United States of America			
State (that is, country) of nationality: US		State (that is, country) of residence: US	
Name and address: (Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)  CURRENS, Michael 615 Wilson Place Frederick, MD 21702 United States of America			
State (that is, country) of nationality: US		State (that is, country) of residence: US	
<input checked="" type="checkbox"/> Further applicants are indicated on a continuation sheet.			

## Continuation of Box No. II

## APPLICANT(S)

*If none of the following sub-boxes is used, this sheet should not be included in the demand.*

Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)*

REIN, Alan  
7295 Swan Point Way  
Columbia, MD 21045  
United States of America

State *(that is, country)* of nationality:

US

State *(that is, country)* of residence:

US

Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)*

FENG, Ya-Xiong  
10300 West Lake Drive, Apt. 305  
Bethesda, MD 20817  
United States of America

State *(that is, country)* of nationality:

US

State *(that is, country)* of residence:

US

Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)*

FISHER, Robert  
17408 Miller's Sawmill Road  
Sharpsburg, MD 21782  
United States of America

State *(that is, country)* of nationality:

US

State *(that is, country)* of residence:

US

Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)*

STEPHEN, Andrew  
114 North Beechwood Avenue  
Catonsville, MD 21228  
United States of America

State *(that is, country)* of nationality:

US

State *(that is, country)* of residence:

US

☒ Further applicants are indicated on another continuation sheet.

## Continuation of Box No. II

## APPLICANT(S)

*If none of the following sub-boxes is used, this sheet should not be included in the demand.*

Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)*

WORTHY, Karen  
13801 Wanegarden Drive  
Germantown, MD 20874  
United States of America

State *(that is, country)* of nationality:

US

State *(that is, country)* of residence:

US

Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)*

SEI, Shizuko  
10104 Galsworthy Place  
Bethesda, MD 20814  
United States of America

State *(that is, country)* of nationality:

JP

State *(that is, country)* of residence:

US

Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)*

CRISE, Bruce  
208 Chestnut Avenue  
Washington Grove, MD 20880  
United States of America

State *(that is, country)* of nationality:

US

State *(that is, country)* of residence:

US

Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)*

HENDERSON, Louis E.  
10296 Quail Trail  
Mt. Airy, MD 21771  
United States of America

State *(that is, country)* of nationality:

US

State *(that is, country)* of residence:

US

☐ Further applicants are indicated on another continuation sheet.

**Box No. III AGENT OR COMMON REPRESENTATIVE; OR ADDRESS FOR CORRESPONDENCE**The following person is ☒ agent ☐ common representativeand ☒ has been appointed earlier and represents the applicant(s) also for international preliminary examination.☐ is hereby appointed and any earlier appointment of (an) agent(s)/common representative is hereby revoked.☐ is hereby appointed, specifically for the procedure before the International Preliminary Examining Authority, in addition to the agent(s)/common representative appointed earlier.Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)*

LOCKYER, Jean M.  
 TOWNSEND AND TOWNSEND AND CREW LLP  
 Two Embarcadero Center, Eighth Floor  
 San Francisco, California 94111-3834  
 United States of America

Telephone No.:

415-576-0200

Facsimile No.:

415-576-0300

Teleprinter No.:

Agent's registration No. with the Office

44,879

☐ **Address for correspondence:** Mark this check-box where no agent or common representative is/has been appointed and the space above is used instead to indicate a special address to which correspondence should be sent.**Box No. IV BASIS FOR INTERNATIONAL PRELIMINARY EXAMINATION****Statement concerning amendments:\***

1. The applicant wishes the international preliminary examination to start on the basis of:

☒ the international application as originally filedthe description ☐ as originally filed☒ as amended under Article 34the claims ☒ as originally filed☐ as amended under Article 19 (together with any accompanying statement)☐ as amended under Article 34the drawings ☒ as originally filed☐ as amended under Article 342. ☐ The applicant wishes any amendment to the claims under Article 19 to be considered as reversed.3. ☐ The applicant wishes the start of the international preliminary examination to be postponed until the expiration of applicable time limit under Rule 69.1(d).

\* Where no check-box is marked, international preliminary examination will start on the basis of the international application as originally filed or, where a copy of amendments to the claims under Article 19 and/or amendments of the international application under Article 34 are received by the International Preliminary Examining Authority before it has begun to draw up a written opinion or the international preliminary examination report, as so amended.

**Language for the purposes of international preliminary examination: ENGLISH**☒ which is the language in which the international application was filed.☐ which is the language of a translation furnished for the purposes of international search.☐ which is the language of publication of the international application.☐ which is the language of the translation (to be) furnished for the purposes of international preliminary examination.**Box No. V ELECTION OF STATES**

The filing of this demand constitutes the election of all Contracting States which are designated and are bound by Chapter II of the PCT.

**Box No. VI CHECK LIST**

The demand is accompanied by the following elements, in the language referred to in Box No. IV, for the purposes of international preliminary examination:

- |  |   |   |        |
|--|---|---|--------|
| 1. translation of international application                              | : |   | sheets |
| 2. amendments under Article 34   | : | 1 | sheets |
| 3. copy (or, where required, translation) of amendments under Article 19 | : |   | sheets |
| 4. copy (or, where required, translation) of statement under Article 19  | : |   | sheets |
| 5. letter  | : | 1 | sheets |
| 6. other ( <i>specify</i> )  | : |   | sheets |

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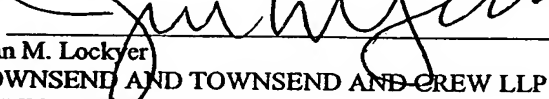
<input type="checkbox"/>	<input type="checkbox"/>
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<input type="checkbox"/>	<input type="checkbox"/>
<input type="checkbox"/>	<input type="checkbox"/>

The demand is also accompanied by the item(s) marked below:

- |  |  |
|--|--|
| 1. <input checked="" type="checkbox"/> fee calculation sheet                               | 5. <input type="checkbox"/> statement explaining lack of signature                           |
| 2. <input type="checkbox"/> original separate power of attorney                            | 6. <input checked="" type="checkbox"/> sequence listing in computer readable form            |
| 3. <input type="checkbox"/> original general power of attorney                             | 7. <input type="checkbox"/> tables in computer readable form related to a sequence listing   |
| 4. <input type="checkbox"/> copy of general power of attorney; reference number, if any: ; | 8. <input checked="" type="checkbox"/> other ( <i>specify</i> ): Transmittal Letter Postcard |

**Box No. VII SIGNATURE OF APPLICANT, AGENT OR COMMON REPRESENTATIVE**

Next to each signature, indicate the name of the person signing and the capacity in which the person signs (if such capacity is not obvious from reading the demand).

X   
 Jean M. Lockyer  
 TOWNSEND AND TOWNSEND AND CREW LLP  
 USPTO Reg. No.: 44,879  
 Applicants' Agent

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1. Date of actual receipt of DEMAND:	
2. Adjusted date of receipt of demand due to CORRECTIONS under Rule 60.1(b):	
3. <input type="checkbox"/> The date of receipt of the demand is AFTER the expiration of 19 months from the priority date and item 4 or 5, below, does not apply. <input type="checkbox"/> The applicant has been informed accordingly.	6. <input type="checkbox"/> The date of receipt of the demand is AFTER the expiration of the time limit under Rule 54bis.1(a) and item 7 or 8, below, does not apply
4. <input type="checkbox"/> The date of receipt of the demand is WITHIN the time limit of 19 months from the priority date as extended by virtue of Rule 80.5.	7. <input type="checkbox"/> The date of receipt of the demand is WITHIN the time limit under Rule 54bis.1(a) as extended by virtue of Rule 80.5.
5. <input type="checkbox"/> Although the date of receipt of the demand is after the expiration of 19 months from the priority date, the delay in arrival is EXCUSED pursuant to Rule 82.	8. <input type="checkbox"/> Although the date of receipt of the demand is after the expiration of the time limit under Rule 54bis.1(a), the delay in arrival is EXCUSED pursuant to Rule 82.

For International Bureau use only

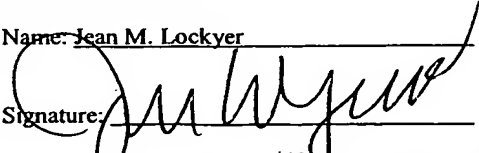
Demand received from IPEA on:

## PCT

## FEE CALCULATION SHEET

## Annex to the Demand

For International Preliminary Examining Authority use only

International application No. <b>PCT/US2003/032086</b> Applicant's or agent's file reference <b>15280-4621PC</b>	Date stamp of the IPEA
Applicant <b>THE GOVERNMENT OF THE USA AS          REPRESENTED BY THE SECRETARY OF THE          DEPT. OF HEALTH AND HUMAN SERVICES et al.</b>	
<b>CALCULATION OF PRESCRIBED FEES</b>  <div style="display: flex; justify-content: space-between;"> <div style="width: 40%;">           1. Preliminary examination fee .....             2. Handling fee (<i>Applicants from certain States are entitled to a reduction of 75% of the handling fee. Where the applicant is (or all applicants are) so entitled, the amount to be entered at H is 25% of the handling fee.</i>) .....             3. Total of prescribed fees            Add the amounts entered at P and H            and enter total in the TOTAL box .....         </div> <div style="width: 55%; border: 1px solid black; padding: 5px;"> <div style="display: flex; justify-content: space-between; align-items: center;"> <span>600.00</span> <span style="border: 1px solid black; padding: 2px 5px;">P</span> </div> <div style="display: flex; justify-content: space-between; align-items: center; margin-top: 10px;"> <span>162.00</span> <span style="border: 1px solid black; padding: 2px 5px;">H</span> </div> <div style="display: flex; justify-content: space-between; align-items: center; margin-top: 10px;"> <span>762.00</span> </div> <div style="display: flex; justify-content: space-between; align-items: center; margin-top: 5px;"> <span>TOTAL</span> </div> </div> </div>	
<b>MODE OF PAYMENT</b>  <div style="display: flex; justify-content: space-between;"> <div style="width: 45%;"> <input checked="" type="checkbox"/> authorization to charge deposit account with the IPEA (see below)   <input type="checkbox"/> cheque   <input type="checkbox"/> postal money order   <input type="checkbox"/> bank draft         </div> <div style="width: 45%;"> <input type="checkbox"/> cash   <input type="checkbox"/> revenue stamps   <input type="checkbox"/> coupons   <input type="checkbox"/> other (<i>specify</i>):         </div> </div>	
<b>AUTHORIZATION TO CHARGE (OR CREDIT) DEPOSIT ACCOUNT</b> <i>(This mode of payment may not be available at all IPEAs)</i>  <input checked="" type="checkbox"/> Authorization to charge the total fees indicated above. <input checked="" type="checkbox"/> <i>(this check-box may be marked only if the conditions for deposit accounts of the IPEA so permit)</i> Authorization to charge any deficiency or credit any overpayment in the total fees indicated above.	IPEA/ <u>US</u>  Deposit Account No.: <u>20-1430</u>  Date: <u>07 May 2004</u>  Name: <u>Jean M. Lockyer</u>  Signature: 

### High-throughput screen

[0084] A high throughput screen was used to detect small molecules that disrupted the association of viral NC-p7 protein with an oligonucleotide. The assay was performed using a Tomtec Quadra robotic system. NC protein was immobilized on Costar (Corning, NY) high-bind polystyrene 96 well plates by incubating 100 µl 250nM NC-p7 in phosphate buffered saline (PBS)/ 10µM zinc chloride/10mM β-mercaptoethanol/0.05% Tween overnight at 4°C. 200µl of 2% BSA in PBS/10µM zinc chloride/10mM β-mercaptoethanol/0.05% Tween was added and incubated at room temperature for 1-2 hours in order to block the plates. The plates were then washed 2 times with 200µl PBS/10µM zinc chloride/10mM β-mercaptoethanol/0.05% Tween using a Titertek M96V plate washer and stored overnight at 4°C. 5nM biotinylated 28 base oligonucleotide (5' GACTTGTGGAAAATCTCTAGC AGTGCAT 3'; SEQ ID NO:1) in PBS/10µM zinc chloride/10mM β-mercaptoethanol/0.05% Tween was added to each well followed by 10µM of test compound (in 20% dimethyl sulphoxide) from the Diversity Set and allowed to incubate at room temperature for 1 hour. Plates were then washed 3 times with 200µl PBS/0.05% Tween. Binding of the biotinylated 28mer was measured by adding 100µl 1:20000 dilution of nutraavidin-horse radish peroxidase (stock 0.8mg/ml, from Pierce Chemical Co. Rockford, Il) and incubating at room temperature for 1 hour. The plates were washed 3 times with 200µl PBS/0.05% Tween. Plates were developed by adding 100µl of Supersignal (Pierce Chemical Co. Rockford, Il) and bioluminescence was measured in a Wallac Victor plate reader. Each plate had eight positive (5mM EDTA in PBS) and eight negative (20% DMSO alone) controls. The hit threshold was set at 100% inhibition. Active compounds that were identified included NSC 13778, NSC 13746, NSC 13755, and NSC 28620.

### Secondary screen

[0085] A secondary screen was used to identify small molecules that disrupt *in vitro* assembly of Gag precursor protein into virus-like particles. Artificial viral particles were assembled from recombinant Gag protein and tRNA as previously described (*see, e.g.,* Campbell & Rein, "In vitro assembly properties of human immunodeficiency virus type 1 Gag protein lacking the p6 domain" *J. Virol.* 73: 2270-2279, 1999).

[0086] Two series of active compounds were identified that disrupted viral assembly. One series, an aromatic antimony-containing series, is based on the antimony-containing

### High-throughput screen

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[0086] Two series of active compounds were identified that disrupted viral assembly. One series, an aromatic antimony-containing series, is based on the antimony-containing

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LLP

Palo Alto,  
Tel 650 326-2400

Denver, Colorado  
Tel 303 571-4000

Walnut Creek, California  
Tel 925 472-5000

Seattle, Washington  
Tel 206 467-9600

10/528747  
JC14 Rec'd PCT/PTO  
22 MAR 2005  
San Francisco

Two Embarcadero Center  
8th Floor  
San Francisco  
California 94111-3834  
Tel 415 576-0200  
Fax 415 576-0300

May 7, 2004

**VIA EXPRESS MAIL**

Special Programs Examiner Sue Wolski  
Mail Stop PCT  
Commissioner for Patents  
P.O. Box 1450  
Alexandria, VA 22313-1450

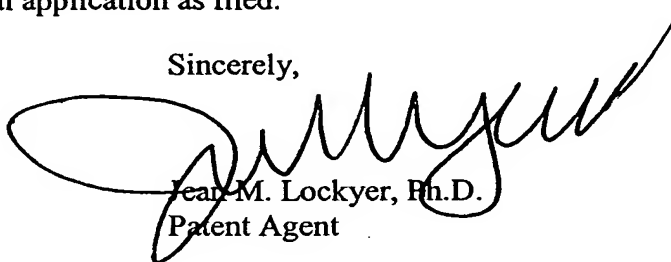
Re: International Application No. PCT/US2003/032086  
filed October 8, 2003  
Applicant(s): The Government of the United States of America *et al.*  
Inventor(s): Shoemaker *et al.*  
Title: Identification of Anti-HIV Compounds Inhibiting Virus Assembly and  
Binding of Nucleocapsid Protein to Nucleic Acid  
Our Ref.: 15280-4621PC

Dear Special Programs Examiner Wolski:

In order to comply with the WIPO Standard ST.25 requirements for the Sequence Listing, please find enclosed the Sequence Listing and disk at this time.

It is hereby stated that "the information recorded on the computer readable form is identical to the written sequence listing" and does not include matter which goes beyond the disclosure in the international application as filed.

Sincerely,



Jean M. Lockyer, Ph.D.  
Patent Agent

JML:dmw  
Enclosures  
60209915 v1

SEQUENCE LISTING

<110> Shoemaker, Robert H.  
Currens, Michael  
Rein, Alan  
Feng, Ya-Xiong  
Fisher, Robert  
Stephen, Andrew  
Worthy, Karen  
Sei, Shizuko  
Crise, Bruce  
Henderson, Louis E.  
The Government of the United States of America  
as represented by The Secretary of the  
Department of Health and Human Services

<120> Identification of Anti-HIV Compounds Inhibiting Virus  
Assembly and Binding of Nucleocapsid Protein to Nucleic  
Acid

<130> 015280-462100PC

<140> WO PCT/US03/32086  
<141> 2003-10-08

<150> US 60/416,854  
<151> 2002-10-08

<160> 1

<170> PatentIn Ver. 2.1

<210> 1  
<211> 28  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Description of Artificial Sequence:biotinylated 28  
base oligonucleotide for viral NC-p7 protein  
association assay

<400> 1  
gacttgtgga aaatctctag cagtgcac

28